



**UNITED STATES AIR FORCE
ARMSTRONG LABORATORY**

**Acute Inhalation Toxicity Evaluation
of a 9:1 Mixture of
1,1,1,3,3,3-Hexafluoropropane
and 1-Bromopropane,
A Replacement Candidate for Ozone
Depleting Substances**

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TECHNICAL REVIEW AND APPROVAL

AL/OE-TR-1997-0061

The animals used in this study were handled in accordance with the principles stated in the *Guide for the Care and Use of Laboratory Animals*, Institute of Laboratory Animal Resources, National Research Council, National Academy Press, 1996, and the Animal Welfare Act of 1966, as amended.

This report has been reviewed by the Office of Public Affairs (PA) and is releasable to the National Technical Information Service (NTIS). At NTIS, it will be available to the general public, including foreign nations.

This technical report has been reviewed and is approved for publication.

FOR THE COMMANDER


TERRY A. CHILDRESS, Lt Col, USAF, BSC
Director, Toxicology Division
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PREFACE

This is one of a series of technical reports describing results of the experimental laboratory programs conducted at the Toxicology Division under the ManTech Geo-Centers Joint Venture Toxic Hazards Research contract. This document serves as a final report on the acute inhalation toxicity of the 9:1 mixture of 1,1,1,3,3,3-Hexafluoropropane and 1-Bromopropane, a replacement candidate for ozone depleting substances. The research described in this report began in November 1996 and was completed in December 1996 under Department of the Air Force Contract No. F41624-96-C-9010. Lt Col Terry A. Childress served as the Contracting Officer's Representative for the U.S. Air Force, Armstrong Laboratory. Darol E. Dodd, Ph.D., served as Program Manager for ManTech Geo-Centers Joint Venture.

The animals used in this study were handled in accordance with the principles stated in the *Guide for the Care and Use of Laboratory Animals*, Institute of Laboratory Animal Resources, National Academy Press, 1996, and the Animal Welfare Act of 1966, as amended. The authors gratefully acknowledge the technical assistance of Marcia Feldmann, Richard J. Godfrey, Jerry W. Nicholson, Margaret A. Parish, and Darol E. Dodd, Ph.D.

ABBREVIATIONS

BP	1-Bromopropane
°C	Degrees Centigrade
CFCs	Chlorofluorocarbons
Conc.	Concentration
DoD	Department of Defense
F-344	Fischer 344 rat(s)
g	Gram(s)
GC	Gas chromatograph
h	Hour(s)
HFC-236fa	1,1,1,3,3,3-Hexafluoropropane
I.D.	Internal diameter
Inj	Injections
L	Liter
LD50	Median lethal dose
M	Meter
m ³	Cubic meter
min	Minute(s)
mg	Milligram
mL	Milliliter
mm	Millimeter
ppm	Parts per million
SD	Standard deviation
µm	Microns

SECTION I

INTRODUCTION

Fire extinguishant agents, refrigerants, and other solvents presently in the Department of Defense (DOD) inventory contain halogenated fluorocarbons. Chloro- and bromofluorocarbons (halons) are substances thought to cause ozone depletion in the stratosphere. Environmental concern over potential ozone depletion by activity of chlorine radicals from chlorofluorocarbons (CFCs) has led to an international treaty called the Montreal Protocol (1987) which calls for the phaseout of select CFCs and halons by the year 2000. The potential utility of a number of chemical substitutes that have little or no ozone depleting potential is being investigated to meet the demand for alternatives to CFCs and halons

The DOD requires the development of a complete toxicity profile for the potential chemical replacements which includes the results of acute toxicity testing. Because these replacements are currently being developed and are not manufactured commercially, very little, if any, toxicity information is available in the literature. To initiate responsible industrial hygiene practice within the production area and provide or recommend appropriate protective equipment in the workplace, it is necessary that operational personnel are aware of the acute health hazards of this compound.

A 9:1 mixture of 1,1,1,3,3,3-hexafluoropropane (HFC-236fa) and 1-bromopropane (BP) was developed by the University of New Mexico as a chemical replacement candidate for ozone depleting fire extinguishants. HFC-236fa, a colorless gas, is a refrigerant developed through a joint EPA/U.S. Navy effort (Vinegar et al., 1996). No acute toxicity data were found in the literature for HFC-236fa. 1-Bromopropane is a toxic (LD_{50} 25,300 mg/m³), highly flammable colorless liquid (IOSHIC, 1989) which is irritating to the skin. Vapors of BP are irritating to the eyes, mucous membranes, and the respiratory tract. Long-term exposure to BP can cause hepatic and renal damage.

The toxicity associated with acute exposure to the 9:1 mixture of HFC-236fa and BP is not known; therefore, an acute inhalation limit test was performed to determine the toxicity associated with acute inhalation exposure to the mixture. The data obtained from this inhalation toxicity test will provide a measure of toxic potency that can be compared with other chemicals, including other CFCs and halon replacement candidates. The species and sex of animals selected for this acute toxicity test were in conformance with the requirements of the U.S. Environmental Protection Agency (1982). Existing alternative methods to animal testing were inadequate for use in this study.

SECTION II

MATERIALS AND METHODS

Test Materials

The 9:1 mixture of HFC-236fa and BP was provided by the University of New Mexico, New Mexico Engineering Research Institute. The mixture was contained in a cylinder as a mixed liquid under pressure. To obtain a representative sample, the mixture was shaken, then opened from the bottom to deliver the liquid into a flexible confined space (1- and 5-L Tedlar sample bags). Pertinent chemical and physical properties of the components are listed below.

1,1,1,3,3,3-Hexafluoropropane

Trade Name:	HFC-236fa
Source:	PCR, Inc. Gainesville, FL
CAS No.:	690-39-1
Boiling Point:	-0.7 °C
Molecular Wt:	152.04
Appearance:	Colorless gas

1-Bromopropane

Source:	Aldrich Chemical Co., Inc. Milwaukee, WI
CAS No.:	106-94-5
Boiling Point:	71 °C
Molecular Weight:	122.99
Vapor pressure:	146 mmHg @ 20 °C
Specific Gravity:	1.354 g/mL
Appearance:	Colorless liquid

No compositional analysis was performed by this laboratory on the mixture as received.

Test Animals

Fischer 344 (F-344) rats (CDF®[F-344]Cr1BR), 7 weeks of age, were purchased from Charles River Breeding Laboratory, Wilmington, MA. All animals were identified by tattoo and subjected to a two-week acclimation period. Rats were group housed (two per cage, separated by sex) in clear plastic cages with hardwood-chip bedding (Sani-Chip®, P.J. Murphy Forest Products, Montville, NJ). Water and feed (Certified Rodent Diet #5002, PMI Feeds, Inc., St Louis, MO) were available *ad libitum*, except for during the 4-h exposure period. Animal room temperatures were maintained at 21 to 25 °C and the light/dark cycle was set at 12-h intervals.

Experimental Design

Acute Inhalation Toxicity Limit Test

Five male and five female F-344 rats were exposed for 4 h to a target concentration of 5 mg/L of the HFC-236fa/BP mixture. Exposures were performed using a nose-only inhalation chamber (Cannon et al., 1983). Animals body weights were recorded prior to exposure and 1, 2, 4, 7, and 14 days postdosing. Animals were observed twice daily during the postexposure period, and any clinical signs of toxic stress (such as chromodacryorrhea, nasal discharge, or abnormal breathing) were recorded. Rats were

euthanatized (CO₂ inhalation) and gross pathology performed on Day 14 postexposure. No further testing of this mixture was performed since no compound-related mortality was observed at the limit test concentration of 5 mg/L.

Exposure Atmosphere Generation and Analysis

At normal room temperature and atmospheric pressure, the 9:1 HFC-236fa and BP mixture was completely vaporized. A 5-L Tedlar sample bag was used to contain the volume of mixture required for the inhalation exposure. The sample bag was pressurized in order to deliver the mixture into the chamber input air stream.. This forced the vapor through a controlling valve, flow meter, and then through a counter current mixing and dilution system. The exposure concentration was controlled in response to analysis of the chamber atmosphere.

The average molecular weight of the mixture was determined (149.1) and then dilution bags were prepared using the calculated values. A Varian 3400 gas chromatograph (GC) equipped with a 15-M x 0.53-mm SPB-5 loop injector (Supleco, Bellefonte, PA; Model 2-5304, Lot #1133120, I.D. 0.53 μ m), flame ionization detector, and an inboard integrator was used to analyze the mixture concentration. The chamber atmosphere was sampled at 5-min intervals every 10 min during the 4-h exposure. Three injections were made during each interval of sampling. Area units were converted to concentration based on a calibration curve. Appendix A contains calibration data for the GC analyses of HFC-236fa and BP. Appendix B contains calibration data for the flow meters used in the exposure atmosphere generation system (M601) and chamber atmosphere analysis (M604).

SECTION IV

RESULTS

Acute Inhalation Toxicity

Five male and five female rats were exposed to the 9:1 HFC-236fa/BP mixture. The mean mixture concentration for the 4-h exposure was 5.09 mg/L (SD 0.17). The distribution of the two components based on area units was 89.5% HFC-236fa and 10.5% BP (Appendix C). No deaths resulted from the acute inhalation exposure, and no signs of toxicity were observed postexposure. All male rats gained weight over the 14-day observation period (Table 1). Two of the five female rats gained weight, whereas the other three females lost weight during the postexposure observation period. No gross lesions were observed at necropsy for any animals on study.

TABLE 1. BODY WEIGHTS^a OF F-344 RATS AFTER ACUTE INHALATION EXPOSURE TO 5 mg/L OF A 9:1 MIXTURE OF HFC-236fa AND BP

Animal Number	Study Day				
	0	1	2	7	14
Male					
01	266.3	263.4	262.6	269.7	273.1
02	284.5	277.8	278.7	280.5	285.8
03	268.3	263.9	259.0	261.8	271.7
04	245.2	238.5	236.0	237.9	252.1
05	289.8	284.7	284.1	290.6	294.9
Mean	270.8	265.7	264.1	268.1	275.5
SD	17.5	17.7	18.9	20.1	16.2
Female					
01	175.8	171.5	173.6	173.6	177.7
02	180.7	176.7	178.4	179.7	183.5
03	171.0	170.2	168.0	169.5	170.9
04	183.5	180.3	178.2	179.0	176.7
05	163.8	156.6	155.4	152.7	154.5
Mean	175.0	171.1	170.7	170.9	172.7
SD	7.9	9.0	9.6	11.0	11.1

^aWeight in grams.

SECTION V

DISCUSSION

In this inhalation toxicity study of the 9:1 mixture of HFC-236fa and BP, no deaths or signs of toxic stress were observed in any of the animals exposed at the limit test value of 5 mg/L. Under the conditions of the limit test performed in this laboratory, this 9:1 mixture of HFC-236fa and BP did not demonstrate an acute toxicological hazard when administered by the inhalation route.

SECTION VI

REFERENCES

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U.S. Environmental Protection Agency. 1982. Health Effects Test Guidelines (Report No. EPA 56016-82-001). Washington, DC: Office of Pesticides and Toxic Substances.

Vinegar, A., G.W. Buttler, M.C. Caracci, and J.D. McCafferty. 1996. Gas uptake kinetics of 1,1,1,3,3,3-hexafluoropropane (HFC-236fa) and identification of its potential metabolites. In: 1995 Toxic Hazards Research Unit Annual Report. D.E. Dodd, ed. AL/OE-TR-1996-0132. Wright-Patterson Air Force Base, OH: Armstrong Laboratory. pp. 78-89.

APPENDIX A

GC Calibration for HFC-236fa/BP Mixture

Concentration (mg/L)	HFC-236fa* Area Units	BP* Area Units	Total Area Units
2.5	339166	40126	379292
2.5	341656	41131	382787
2.5	342393	45618	388011
5.0	692258	89221	781479
5.0	652810	82359	735169
5.0	737187	93462	830649
7.5	1008097	123310	1131407
7.5	1033638	121774	1155412
7.5	1049480	129795	1179275
<hr/>			
	% of total	% of total	
2.5	89.42081	10.57918	
2.5	89.25485	10.74514	
2.5	88.24311	11.75688	
5.0	88.58305	11.41694	
5.0	88.79726	11.20273	
5.0	88.74831	11.25168	
7.5	89.10118	10.89881	
7.5	89.46055	10.53944	
7.5	88.99366	11.00633	
<hr/>			
Mean %	88.95587	11.04412	

*Each value represents the sum of three injections.

Appendix B

Flow Meter Calibrations

M601 Flow Meter HFC-236fa/BP Mixture

Glass Bead	
Reading	mL/min
10	7.49
20	9.66
30	13.83

Calibration Data for M604 Flow Meter

Air			
Inches of Water	Glass Bead Reading	Stainless Steel Bead Reading	L/min
0.06	40	20	2.41
		20	2.40
		20	2.38
1.8	80	40	4.80
		40	4.78
		40	4.80
3.5	118	60	7.07
		60	7.09
		60	7.10
5.3	reading off scale	80	9.46
		80	9.49
		80	9.48
5.8	---	85	10.01
		85	10.00
		85	10.02
7.2	---	100	11.79
		100	11.78
		100	11.75
8.8	---	120	14.07
		120	14.07
		120	14.04

APPENDIX C

Exposure Data 19 Nov 96

90% 1,1,1,3,3,3-Hexafluoropropane/10% 1-Bromopropane Mixture

Target Concentration 5 mg/L

Time	HFC-236fa Area Units	BP Area Units	TOTAL Area Units	Area Units Sum of 3 Inj	Conc. mg/L	HFC-236fa % of total	BP % of total
	82013	8984	90997			90.1	9.9
	281420	29948	311368			90.4	9.6
	270835	33205	304040	706405	4.56	89.1	10.9
	277424	36625	314049			88.3	11.7
	275402	37634	313036			88.0	12.0
	261569	34821	296390	923475	5.97	88.3	11.7
	265363	33019	298382			88.9	11.1
	262121	31835	293956			89.2	10.8
	261211	31284	292495	884833	5.72	89.3	10.7
Begin							
8:45	259304	30620	289924			89.4	10.6
	257082	30174	287256			89.5	10.5
	255570	29863	285433	862613	5.58	89.5	10.5
	253726	29584	283310			89.6	10.4
	264797	30824	295621			89.6	10.4
	250849	29271	280120	859051	5.55	89.6	10.4
	248309	28942	277251			89.6	10.4
	246651	28787	275438			89.5	10.5
	246150	28634	274784	827473	5.35	89.6	10.4
	243883	28351	272234			89.6	10.4
	243453	28277	271730			89.6	10.4
	241720	28117	269837	813801	5.26	89.6	10.4
	240210	27982	268192			89.6	10.4
	238716	27699	266415			89.6	10.4
	238356	27670	266026	800633	5.17	89.6	10.4
	236305	27128	263433			89.7	10.3

Appendix C cont'd

	HFC- 236fa Area Units	BP Area Units	TOTAL Area Units	Area Units Sum of 3 Inj	Conc. mg/L	HFC- 236fa % of total	BP % of total
	234223	27125	261348			89.6	10.4
	233804	26906	260710	785491	5.08	89.7	10.3
	240617	27856	268473			89.6	10.4
	236660	27446	264106			89.6	10.4
	234869	27330	262199	794778	5.14	89.6	10.4
	232931	27096	260027			89.6	10.4
	232284	27032	259316			89.6	10.4
	231080	26800	257880	777223	5.02	89.6	10.4
	230667	26758	257425			89.6	10.4
	228405	26533	254938			89.6	10.4
	227650	26453	254103	766466	4.95	89.6	10.4
	241726	28028	269754			89.6	10.4
	239410	27868	267278			89.6	10.4
	249107	29028	278135	815167	5.27	89.6	10.4
	247023	28928	275951			89.5	10.5
	245290	28688	273978			89.5	10.5
	243664	28584	272248	822177	5.31	89.5	10.5
	241724	28226	269950			89.5	10.5
	243974	28476	272450			89.5	10.5
	241747	28191	269938	812338	5.25	89.6	10.4
	239689	27875	267564			89.6	10.4
	238691	27863	266554			89.5	10.5
	237957	27766	265723	799841	5.17	89.6	10.4
9:45	236801	27715	264516			89.5	10.5
	236117	27614	263731			89.5	10.5
	235633	27565	263198	791445	5.11	89.5	10.5
	235062	27457	262519			89.5	10.5
	233022	27167	260189			89.6	10.4
	232796	27014	259810	782518	5.06	89.6	10.4
	232648	27151	259799			89.5	10.5
	231886	27115	259001			89.5	10.5
	230132	26749	256881	775681	5.01	89.6	10.4
	228365	26491	254856			89.6	10.4
	227862	26602	254464			89.5	10.5
	226688	26440	253128	762448	4.93	89.6	10.4
	226384	26325	252709			89.6	10.4
	226600	26404	253004			89.6	10.4

Appendix C cont'd

	HFC- 236fa Area Units	BP Area Units	TOTAL Area Units	Area Units Sum of 3 Inj	Conc. mg/L	HFC- 236fa % of total	BP % of total
	225641	26317	251958	757671	4.90	89.6	10.4
	224587	26180	250767			89.6	10.4
	223375	25855	249230			89.6	10.4
	245188	28736	273924	773921	5.00	89.5	10.5
	243945	28553	272498			89.5	10.5
	243111	28536	271647			89.5	10.5
	241862	28395	270257	814402	5.26	89.5	10.5
	241526	28196	269722			89.5	10.5
	240612	28111	268723			89.5	10.5
	239758	28052	267810	806255	5.21	89.5	10.5
	240175	28210	268385			89.5	10.5
	250644	29232	279876			89.6	10.4
	238948	28024	266972	815233	5.27	89.5	10.5
	228476	26791	255267			89.5	10.5
	229588	27031	256619			89.5	10.5
	222858	25922	248780	760666	4.92	89.6	10.4
	221396	25879	247275			89.5	10.5
	226304	26428	252732			89.5	10.5
	224750	26399	251149	751156	4.85	89.5	10.5
	223323	26256	249579			89.5	10.5
	221868	26104	247972			89.5	10.5
	226245	26503	252748	750299	4.85	89.5	10.5
10:45	225128	26429	251557			89.5	10.5
	224073	26348	250421			89.5	10.5
	223100	26106	249206	751184	4.85	89.5	10.5
	222366	26109	248475			89.5	10.5
	220954	25961	246915			89.5	10.5
	221620	26084	247704	743094	4.80	89.5	10.5
	220451	25719	246170			89.6	10.4
	230332	27009	257341			89.5	10.5
	229378	27025	256403	759914	4.91	89.5	10.5
	228693	26959	255652			89.5	10.5
	227546	26754	254300			89.5	10.5
	226791	26635	253426	763378	4.93	89.5	10.5
	225905	26554	252459			89.5	10.5
	225285	26391	251676			89.5	10.5
	224077	26251	250328	754463	4.88	89.5	10.5
	223128	26235	249363			89.5	10.5

Appendix C cont'd

	HFC- 236fa Area Units	BP Area Units	TOTAL Area Units	Area Units Sum of 3 Inj	Conc. mg/L	HFC- 236fa % of total	BP % of total
	222058	26080	248138			89.5	10.5
	221628	26027	247655	745156	4.82	89.5	10.5
	221244	25924	247168			89.5	10.5
	221325	25844	247169			89.5	10.5
	221159	25845	247004	741341	4.79	89.5	10.5
	240445	28269	268714			89.5	10.5
	239800	28111	267911			89.5	10.5
	239697	28061	267758	804383	5.20	89.5	10.5
	237585	27936	265521			89.5	10.5
	238472	28008	266480			89.5	10.5
	237913	27983	265896	797897	5.16	89.5	10.5
	237982	27954	265936			89.5	10.5
	237304	27953	265257			89.5	10.5
	237142	27768	264910	796103	5.14	89.5	10.5
	236551	27833	264384			89.5	10.5
	236361	27711	264072			89.5	10.5
	236399	27781	264180	792636	5.12	89.5	10.5
	236193	27777	263970			89.5	10.5
	235969	27707	263676			89.5	10.5
	235792	27556	263348	790994	5.11	89.5	10.5
11:45	235415	27694	263109			89.5	10.5
	235473	27666	263139			89.5	10.5
	234909	27730	262639	788887	5.10	89.4	10.6
	234936	27757	262693			89.4	10.6
	235009	27701	262710			89.5	10.5
	234616	27609	262225	787628	5.09	89.5	10.5
	234371	27629	262000			89.5	10.5
	234049	27469	261518			89.5	10.5
	234185	27486	261671	785189	5.07	89.5	10.5
	232893	27358	260251			89.5	10.5
	232875	27375	260250			89.5	10.5
	232772	27224	259996	780497	5.04	89.5	10.5
	232483	27254	259737			89.5	10.5
	231582	27214	258796			89.5	10.5
	233958	27537	261495	780028	5.04	89.5	10.5
	239774	28199	267973			89.5	10.5
	239453	28241	267694			89.5	10.5
	239334	28118	267452	803119	5.19	89.5	10.5

Appendix C cont'd

HFC- 236fa Area Units	BP Area Units	TOTAL Area Units	Area Units Sum of 3 Inj	Conc. mg/L	HFC- 236fa % of total	BP % of total
239654	28058	267712			89.5	10.5
239223	28244	267467			89.4	10.6
239096	27988	267084	802263	5.18	89.5	10.5
239796	28260	268056			89.5	10.5
239104	28130	267234			89.5	10.5
238209	28097	266306	801596	5.18	89.4	10.6
238487	28029	266516			89.5	10.5
238310	28161	266471			89.4	10.6
238033	28076	266109	799096	5.16	89.4	10.6
238361	28013	266374			89.5	10.5
238271	28088	266359			89.5	10.5
238794	28172	266966	799699	5.17	89.4	10.6
238111	28017	266128			89.5	10.5
237854	27953	265807			89.5	10.5
236428	27773	264201	796136	5.15	89.5	10.5
235252	27753	263005			89.4	10.6
234099	27595	261694			89.5	10.5
233521	27421	260942	785641	5.08	89.5	10.5
12:45						
End						
MEAN				5.09		
SD				0.17		